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An Encyclopedic Review on Mucoadhesive Drug Delivery System

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ABSTRACT :
Drug actions can be improved by developing new drug delivery systems, such as the mucoadhesive system. These systems remain in close contact with the absorption tissue, the mucous membrane, releasing the drug at the action site leading to a bioavailability increase and both local and systemic effects. Mucoadhesion is currently explained by six theories: electronic, adsorption, wettability, diffusion, fracture and mechanical. Mucoadhesive drug delivery systems interact with the mucus layer covering the mucosal epithelial surface, and mucin molecules increase the residence time of the dosage form at the site of absorption. Bioadhesion may be defined as the state in which two materials, at least one of which is of a biological nature, are held together for extend periods of time by interfacial forces Mucosal layer represents potential sites for the attachment of any bioadhesive systems because mucosal layer lines number of the body including the gastro intestinal tract, the urogenital tract, vaginal tract, the eye, ear, and nose. Buccal route of administration has many advantages such as improving patient compliance, bypassing the GIT and hepatic first pass effect. The mucoadhesive Bilayer tablets consisting of two various types of drug molecules and they show on set of actions at their particular sites.

Keywords: Mucoadhesive drug delivery, Bilayer tablets, polymers, bioadhesive.

I. INTRODUCTION:

The effect of a drug can now be reinforced as a result of the development of new release systems. Controlled release consists of techniques that make the active chemical agents available for a target, providing an adequate release rate and duration to produce the desired effect. The main controlled drug delivery systems currently available include matrices, pellets, floating systems, liposomes, microemulsions, liquid crystals, solid dispersions, nanosuspensions, transdermal systems, cyclodextrin inclusion complexes, osmotic pumps and bioadhesive systems. Bioadhesion can be defined as the state in which two materials, at least one of which is biological in nature, are maintained together for a prolonged time period by means of interfacial forces. During the 1980s, this concept began to be applied to drug delivery systems. It consists of the incorporation of adhesive molecules into some kind of pharmaceutical formulation intended to stay in close contact with the absorption tissue, releasing the drug near to the action site, thereby increasing its bioavailability and promoting local or systemic effects. An extensive review on mucoadhesive systems was compiled by Andrews, Laverty and Jones (2008)[9].

Adhesion can be defined as the bond produced by contact between a pressure sensitive adhesive and a surface. The American Society of Testing and Materials has defined it as the state in which two surfaces are held together by interfacial forces, which may consist of valence forces, interlocking action or both. Mucoadhesive drug delivery systems prolong the residence time of the dosage form at the site of application or absorption[8]. They facilitate an intimate contact of the dosage form with the underlying absorption surface and thus improve the therapeutic performance of the drug. Dosage forms designed for mucoadhesive drug delivery should be small and flexible enough to be acceptable for patients and should not cause irritation. Other desired characteristics of a mucoadhesive dosage form include high drug loading capacity, controlled drug release (preferably unidirectional release), good mucoadhesive properties, smooth surface, tastelessness, and convenient application. Erodible formulations can be beneficial because they do not require system retrieval at the end of desired dosing interval. A number of relevant mucoadhesive dosage forms have been developed for a variety of drugs. Several peptides, including thyrotropin-releasing hormone (TRH), insulin, octreotide, leuprolide, and oxytocin, have been delivered via

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the mucosal route, albeit with relatively low bioavailability (0.1–5%),[3] owing to their hydrophilicity and large molecular weight, as well as the inherent permeation and enzymatic barriers of the mucosa[10].

The potential use for mucoadhesive systems as drug carriers lies in its prolongation of the residence time at the absorption site, allowing intensified contact with the epithelial barrier. On the other hand, adhesion of preparations onto mucous membrane can be impaired by the mucociliary clearance system. This clearance, a natural defense mechanism of the body against the deposition of impurities onto the mucous membrane, can also remove the preparation. Thus, by using bioadhesive molecules, it is possible to retain the preparation at the action site and to direct the drug to a specific site or tissue. Other features associated with the development of controlled drug delivery systems using bioadhesive molecules include a decrease in drug administration frequency and an increase in patient compliance to the therapy . Therefore, a bioadhesive system controlling drug release could improve the treatment of diseases, helping to maintain an effective concentration of the drug at the action site[1].

Mucous membrane is the main administration site for bioadhesive systems, although the need for new bioadhesive formulations for dermal administration has also been reported when prolonged cutaneous action is desired. A prolonged effect upon the dermal administration of creams, solutions, and lotions is unexpected, since such preparations can be easily removed from the skin by moisture, temperature, and physical movements[2] .

Mucousal membranes of human organism are relatively permeable and allow fast drug absorption. They are characterized by an epithelial layer whose surface is covered by mucus. The mucus contains glycoproteins, lipids, inorganic salts and 95% water by mass, making it a highly hydrated system. Mucin is the most important glycoprotein of mucus and is responsible for its structure. The main functions of mucus are protecting and lubricating the epithelium and other additional functions depending on the epithelium covered. Mucus thickness can vary from 50-450 µm in the stomach to less than 1 µm in the oral cavity . The mucous site most used for drug administration and absorption is gastrointestinal, but other routes, including nasal, ocular, buccal, vaginal, rectal, oral, and periodontal have also been studied[3].

This approach to confer Bioadhesion properties has been widely applied in the development of a number of drug delivery systems. Solid micro- and nano-particulate systems based on chitosan and derivatives have been the focus of several studies : microemulsions are thermodynamically stable and isotropic liquid systems, which allow the incorporation of bioadhesive molecules, such as polycarbophil colloidal dispersions of bioadhesive polymers frequently used in preparations for oral hygiene semi-solid systems, as liquid crystalline mesophases and hydrogels which can increase the contact time between preparation and mucous membrane after they undergo in situ gelation. There are a number of materials used for developing such systems[6]. The most studied materials are the polymers derived from polyacrylic acid, such as polycarbophil and carbomers, polymers derived from cellulose, such as hydroxyethylcellulose and carboxymethylcellulose, alginates, chitosan and derivatives and more recently, lectins and their derivatives[5].

Although studies on the mechanisms involved in mucoadhesion and the development of novel mucoadhesive systems and polymers have evolved over the last twenty years, mucoadhesion is not yet fully understood. Quantitative and qualitative techniques are still treated separately. The aim of this study was to systematically review the mechanisms and theories involving mucoadhesion, as well as to describe the methods and polymers most used in mucoadhesive systems for drug delivery[7].

ADVANTAGES OF MUCOADHESIVE DRUG DELIVERY SYSTEM

- Mucoadhesive delivery systems offer several advantages over other oral controlled release systems by virtue of prolongation of residence time of drug in gastrointestinal tract (GIT).
- Targeting and localization of the dosage form at a specific site
- Also, the mucoadhesive systems are known to provide intimate contact between dosage form and the absorptive mucosa, resulting in high drug flux at the absorbing tissue[20].

Mucus Membranes

Mucus membranes (mucosae) are the moist surfaces lining the walls of various body cavities such as the gastrointestinal and respiratory tracts. They consist of a connective tissue layer (the lamina propria) above which is an epithelial layer,

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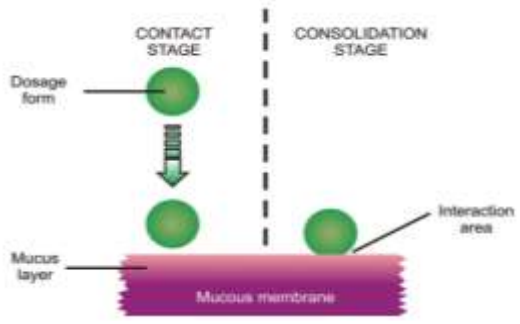
the surface of which is made moist usually by the presence of a mucus layer[19]. The epithelia may be either single layered (e.g. the stomach, small and large intestines and bronchi) or multilayered/stratified (e.g. in the esophagus, vagina and cornea)[18]. The former contain goblet cells which secrete mucus directly onto the epithelial surfaces; the latter contain, or are adjacent to tissues containing, specialized glands such as salivary glands that secrete mucus onto the epithelial surface. Mucus is present either as a gel layer adherent to the mucosal surface or as a luminal soluble or suspended form. The major components of all mucus gels are mucin glycoproteins, lipids, inorganic salts and water, the latter accounting for more than 95% of their weight, making them a highly hydrated system. The major functions of mucus are that of protection and lubrication[17].

MECHANISMS OF MUCOADHESION

The mechanism of adhesion of certain macromolecules to the surface of a mucous tissue is not well understood yet. The mucoadhesive must spread over the substrate to initiate close contact and increase surface contact, promoting the diffusion of its chains within the mucus. Attraction and repulsion forces arise and, for a mucoadhesive to be successful, the attraction forces must dominate[16]. Each step can be facilitated by the nature of the dosage form and how it is administered. For example, a partially hydrated polymer can be adsorbed by the substrate because of the attraction by the surface water[11].



Thus, the mechanism of mucoadhesion is generally divided in two steps, the contact stage and the consolidation stage . The first stage is characterized by the contact between the mucoadhesive and the mucous membrane, with spreading and swelling of the formulation, initiating its deep contact with the mucus layer . In some cases, such as for ocular or vaginal formulations, the delivery system is mechanically attached over the membrane. In other cases, the deposition is promoted by the aerodynamics of the organ to which the system is administered, such as for the nasal route[15]. On the other hand, in the gastrointestinal tract direct formulation attachment over the mucous membrane is not feasible. Peristaltic motions can contribute to this contact, but there is little evidence in the literature showing appropriate adhesion. Additionally, an undesirable adhesion in the esophagus can occur. In these cases, mucoadhesion can be explained by peristalsis, the motion of organic fluids in the organ cavity, or by Brownian motion. If the particle approaches the mucous surface, it will come into contact with repulsive forces (osmotic pressure, electrostatic repulsion, etc.) and attractive forces (van der Waals forces and electrostatic attraction). Therefore, the particle must overcome this repulsive barrier[12].

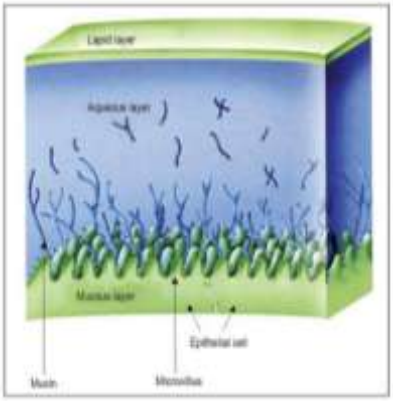


Fig: Mucus membrane structure

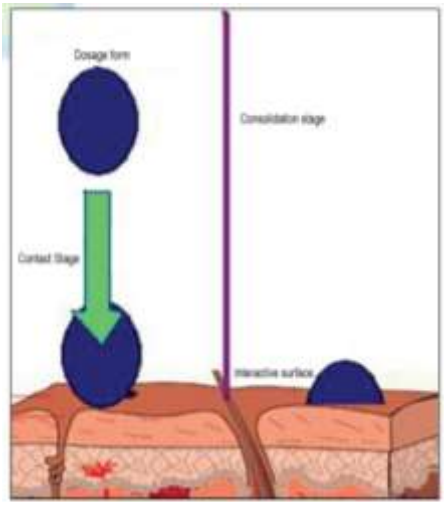


Fig: The process of contact and consolidation

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In the consolidation step the mucoadhesive materials are activated by the presence of moisture. Moisture plasticizes the system, allowing the mucoadhesive molecules to break free and to link up by weak van der Waals and hydrogen bonds .Essentially, there are two theories explaining the consolidation step: the diffusion theory and the dehydration theory[16.13]. According to diffusion theory, the mucoadhesive molecules and the glycoproteins of the mucus mutually interact by means of interpenetration of their chains and the building of secondary bonds. For this to take place the mucoadhesive device has features favoring both chemical and mechanical interactions. For example, molecules with hydrogen bonds building groups (–OH, –COOH), with an anionic surface charge, high molecular weight, flexible chains and surface-active properties, which induct its spread throughout the mucus layer, can present mucoadhesive properties[14] . According to dehydration theory, materials that are able to readily gelify in an aqueous environment, when placed in contact with the mucus can cause its dehydration due to the difference of osmotic pressure. The difference in concentration gradient draws the water into the formulation until the osmotic balance is reached. This process leads to the mixture of formulation and mucus and can thus increase contact time with the mucous membrane. Therefore, it is the water motion that leads to the consolidation of the adhesive bond, and not the interpenetration of macromolecular chains. However, the dehydration theory is not applicable for solid formulations or highly hydrated forms[21].

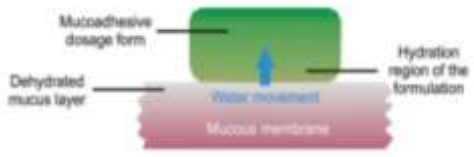

Fig :Dehydration theory of mucoadhesion.

Theories of Mucoadhesion:

Mucoadhesion is a complex process and numerous theories have been proposed to explain the mechanisms involved. These theories include mechanical interlocking, electrostatic, diffusion interpenetration, adsorption and fracture processes.

Wetting Theory

The wetting theory applies to liquid systems which present affinity to the surface in order to spread over it. This affinity can be found by using measuring techniques such as the contact angle. The general rule states that the lower the contact angle, the greater is the affinity. The contact angle should be equal or close to zero to provide adequate spreadability. The spreadability coefficient, S_{AB}, can be calculated from the difference between the surface energies γ_B and γ_A and the interfacial energy γAB, as indicated in the equation given below. This theory explains the importance of contact angle and reduction of surface and interfacial energies to achieve good amount of mucoadhesion[30].

$$S_{AB} = \gamma_B - \gamma_A - \gamma_{AB}$$

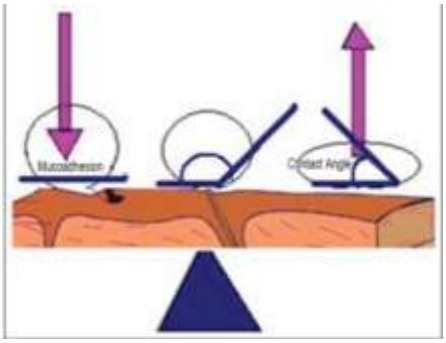

Fig: Influence of contact angle on mucoadhesion

Diffusion Theory :

Diffusion theory describes the interpenetration of both polymer and mucin chains to a sufficient depth to create a semi-permanent adhesive bond . It is believed that the adhesion force increases with the degree of penetration of the polymer chains. This penetration rate depends on the diffusion coefficient, flexibility and nature of the mucoadhesive chains, mobility and contact time. According to the literature, the depth of interpenetration required to produce an efficient bioadhesive bond lies in the range 0.2–0.5 μm[22]. This interpenetration depth of polymer and mucin chains can be estimated by the following equation:

$$l = (tD_b)^{½}$$

where t is the contact time and D_b is the diffusion coefficient of the mucoadhesive material in the mucus. The adhesion strength for a polymer is reached when the depth of penetration is approximately equivalent to the polymer chain size. In order for diffusion to occur, it is important that the components involved have good mutual solubility, that is, both the bioadhesive and the mucus have similar chemical structures. The greater the structural similarity, the better is the mucoadhesive bond[29].

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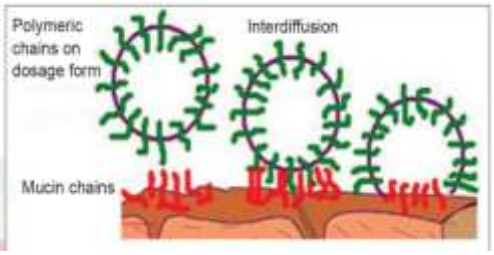

Fig: Secondary interaction between mucoadhesive device and of mucus

Fracture Theory

This is perhaps the most used theory in studies on the mechanical measurement of mucoadhesion. It analyzes the force required to separate two surfaces after adhesion is established. This force, s_m, is frequently calculated in tests of resistance to rupture by the ratio of the maximal detachment force, F_m, and the total surface area, A_0 , involved in the adhesive interaction[23].

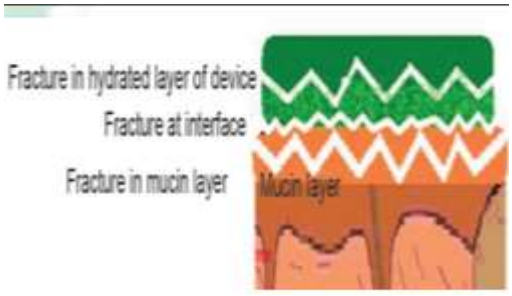

Fig :Fractures occurring for mucoadhesion

Since the fracture theory is concerned only with the force required to separate the parts, it does not take into account the interpenetration or diffusion of polymer chains. Consequently, it is appropriate for use in the calculations for rigid or semi-rigid bioadhesive materials, in which the polymer chains do not penetrate into the mucus layer[28].

The Electronic Theory

This theory describes adhesion occurring by means of electron transfer between the mucus and the mucoadhesive system, arising through differences in their electronic structures. The electron transfer between the mucus and the mucoadhesive results in the formation of double layer of electrical charges at the mucus and mucoadhesive interface. The net result of such a process is the formation of attractive forces within this double layer[27].

The Adsorption Theory

In this instance, adhesion is the result of various surface interactions (primary and secondary bonding) between the adhesive polymer and mucus substrate. Primary bonds due to chemisorptions result in adhesion due to ionic, covalent and metallic bonding, which is generally undesirable due to their permanency.Secondary bonds arise mainly due to van der Waals forces, hydrophobic interactions and hydrogen bonding. Whilst these interactions require less energy to "break", they are the most prominent form of surface interaction in mucoadhesion processes as they have the advantage of being semi-permanent bonds[24].

All these numerous theories should be considered as supplementary processes involved in the different stages of the mucus/substrate interaction, rather than individual and alternative theories. Each and every theory is equally important to describe the mucoadhesion process. There is a possibility that there will be initial wetting of the mucin, and then diffusion of the polymer into mucin layer, thus causing the fracture in the layers to effect the adhesion or electronic transfer or simple adsorption phenomenon that finally leads to the perfect mucoadhesion. The mechanism by which a mucoadhesive bond is formed will depend on the nature of the mucus membrane and mucoadhesive material, the type of formulation, the attachment process and the subsequent environment of the bond. It is apparent that a single mechanism for mucoadhesion proposed in many texts is unlikely for all the different occasions when adhesion occurs[25].

Mechanical Theory

Mechanical theory considers adhesion to be due to the filling of the irregularities on a rough surface by a mucoadhesive liquid. Moreover, such roughness increases the interfacial area available to interactions thereby aiding dissipating energy and can be considered the most important phenomenon of the process. It is unlikely that the mucoadhesion process is the same for all cases and therefore it cannot be described by a single theory.

In fact, all theories are relevant to identify the important process variables. The mechanisms governing mucoadhesion are also determined by the intrinsic properties of the formulation and by the environment in which it is applied. Intrinsic factors of the polymer are related to its molecular weight, concentration and chain flexibility. For linear polymers, mucoadhesion increases with molecular weight, but the same relationship does

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not hold for nonlinear polymers. It has been shown that more concentrated mucoadhesive dispersions are retained on the mucous membrane for longer periods, as in the case of systems formed by in situ gelification[26].

After application, such systems spread easily, since they present rheological properties of a liquid, but gelify as they come into contact the absorption site, thus preventing their rapid removal. Chain flexibility is critical to consolidate the interpenetration between formulation and mucus.

Environment-related factors include pH, initial contact time, swelling and physiological variations. The pH can influence the formation of ionizable groups in polymers as well as the formation of charges on the mucus surface. Contact time between mucoadhesive and mucus layer determines the extent of chain interpenetration. Super-hydration of the system can lead to build up of mucilage without adhesion. The thickness of the mucus layer can vary from 50 to 450 µm in the stomach to less than 1µm in the oral cavity. Other physiological variations can also occur with diseases. None of these mechanisms or theories alone can explain the mucoadhesion which occurs in an array of different situations. However, the understanding of these mechanisms in each instance can help toward the development of new mucoadhesive products[40].

MUCOADHESIVE MATERIALS

The first study presenting the use of a mucoadhesive material was conducted by Nagai, and proposed an improved treatment for stomatitis by using adhesive tablets. Additionally, an increase in the systemic bioavailability of insulin was observed in the form of bioadhesive powder after nasal administration in dogs. Thereafter, bioadhesive materials have been used as absorption promoters for several administration routes. Earlier experiments were also done with known polymers available on the market, such as polyacrylic acids. Currently, the latest research is seeking to develop materials that direct the formulation more specifically to the action site and that can offer other functions besides mucoadhesion such as control over permeation within epithelial tissues, and inactivation of enzymes which can compromise release system action[31].

First Generation Mucoadhesive Materials

These materials are natural or synthetic hydrophilic molecules containing numerous organic functions that generate hydrogen bonds such as carboxyl, hydroxyl and amino groups, which do not adhere specifically onto several surfaces. The very first use of mucoadhesive was as denture fixers and the most known examples are carbomers, chitosans, alginates and cellulose derivatives. They can be incorporated into solid formulations, such as tablets, transdermal adhesives and microparticles, and into semisolid formulations including gels, ointments, pastes and suppositories . These polymers can be subdivided into three classes: cationic, anionic and nonionic

Cationic molecules can interact with the mucus surface, since it is negatively charged at physiological pH. Mucoadhesion of cationic polymers such as chitosan, occurs because of the electrostatic interactions of their amino groups with the sialic groups of mucin in the mucus layer. Chitosan is a semi-synthetic polymer obtained by the deacetylation of chitin and has been extensively investigated as a drug delivery mucoadhesive system. Studies have demonstrated that chitosan can promote the absorption of hydrophilic molecules by the structural reorganization of the proteins associated to the intercellular junctions. The presence of chitosan at the surface of nanoparticles clearly increased their intestinal mucoadhesive behavior in rats. demonstrated in their studies that chitosan showed higher mucoadhesion than carboxymethylcellulose and polycarbophil[39].

In contrast, synthetic polymers derived from polyacrylic acid (carbomers) are negatively charged but are also mucoadhesive. In this case, mucoadhesion results from physical-chemical processes, such as hydrophobic interactions, hydrogen and van der Waals bonds, which are controlled by pH and ionic composition. Polyacrylic acid hydrogels have been extensively studied as mucoadhesive systems. Their chains are flexible and have non-abrasive characteristics when in the partially hydrated state, which decreases the tissue damage caused by friction when they come into contact. The majority of polyacrylic acid derivatives are not water soluble, such as polycarbophil, but form viscous gels when hydrated. Other examples of anionic polymers are carboxymethylcellulose and alginates. The alginates, negatively charged polysaccharides, are widely used in the production of microparticles and are frequently reported as polyanionic mucoadhesive polymers.

Nonionic polymers, including hydroxypropylmethylcellulose, hydroxyethylcellulose and methylcellulose, present

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weaker mucoadhesion force compared to anionic polymers. There is a new class of substances being identified as bioadhesive. This class consists of ester groups of fatty acids, such as glyceryl monooleate and glyceryl monolinoleate, able to build liquid crystals which in turn can act as controlled release systems. These fatty acids build lyotropic liquid crystalline mesophases in the presence of water at body temperature. Liquid crystals can be considered structures of micelles ordered in a molecular arrangement characterized by alternate hydrophobic and hydrophilic regions. Different liquid-crystalline forms including lamellar, hexagonal, and cubic can be built as the surfactant concentration increases. Cubic phase favors the controlled release of drugs, since it has a structure made up of tridimensionalcurved lipid bilayers, separated by congruent water channels. This structure has the appearance of highly viscous transparent gel. Due to this relatively high viscosity, it is difficult to administer on any mucous membrane. In order to circumvent the administration problems, a less viscous mesophase, e.g., the lamellar phase, can be used. In these instances this phase is considered a precursor of the cubic phase. In the case of lyotropic mesophases, the precursor absorbs water in situ and spontaneously builds the cubic phase[38].

Some hydrogels do not build liquid crystals but are able to gelify in situ after exposure to an external stimulus. These are the so-called environmental sensitive polymers and are classified as thermosensitive, e.g. poloxamers and carbomers, pH sensitive, e.g. polyacrylic acid, presenting increased viscosity at higher pH values, glucose sensitive, e.g. polymers linked to concavalin A, electric signal sensitive e.g. polymethacrylic acid, light sensitive, like hyaluronic acid or ionic concentration sensitive, such as gellan gum. All these stimuli are found in the organism, making these polymers of great potential for use in the design of controlled release systems.

Mucoadhesion for gels formed by both liquid crystals and by environmental sensitive polymers can be explained by their rheological properties. These properties decrease the mucociliar clearance and increase the contact time of the formulation with the mucous membrane[32].

Second Generation Mucoadhesive Materials

Studies on novel mucoadhesive systems involve the use of multifunctional materials. An ideal polymer should exhibit the ability to incorporate both hydrophilic and lipophilic drugs, show mucoadhesive properties in its solid and liquid forms, inhibit local enzymes or promote absorption, be specific for a particular cellular area or site, stimulate endocytosis and finally to have a broad safety range .

These novel multifunctional mucoadhesive systems are classified as second generation polymers. They are an alternative to non-specific bioadhesives because they bind or adhere to specific chemical structures on the cell or mucus surface. Good examples of these molecules are lectins, invasins, fimbrial proteins, antibodies , and those obtained by the addition of thiol groups to known molecules.

Lectins are immunogenic vegetal glycoproteins that specifically recognize sugar molecules. They are able to non-covalently bind to glycosilated components of the cellular membrane but not of the mucus, and adhesion can therefore be called cytoadhesion. Through the transmission of a cellular signal, this specific bond can result not only in bioadhesion but also in cellular internalization by different lysosomal and non-lysosomal mechanisms. The most commonly found lectins are those isolated from Abrus precatroius, Agaricus bisporus, Anguilla anguilla, Arachis hypogaea, Pandeiraea simplicifolia, and Bauhinia purpurea[33] .

Bacterial invasins are proteins from the membrane of Yersinia pseudotuberculosis that stimulate fagocytosis at cellular membrane through linkage with integrin receptors. Bacterial fimbrial proteins are able to adhere to the epithelial surface of erythrocytes. This adhesion is related to the pathogenicity of the bacteria. Bacterial adhesive factors can be an efficient mechanism of improving adhesion of mucoadhesive agents used in release systems.

Antibodies can be produced against selected molecules present on the mucus surface. Due to their high specificity, antibodies can be a rational choice as polymeric ligand in the development of site-specific mucoadhesives. This strategy can be useful for instance, in drugs targeting tumor tissues. Thiolated polymers are obtained by the addition of conjugated sulfidryl groups showed that thiolated chitosan increased mucoadhesive properties due to formation of disulfide bridges with cystein domains of glycoproteins of the mucus. Additionally, these products promoted mucus permeation by a mechanism of glutathione regeneration. Finally, they possess antiprotease activity due to their binding ability with divalent cations, such as zinc

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and magnesium, which are co-factors for many proteases. All these characteristics make thiolated chitosan a promising material for administering peptides and proteins in mucous membrane . Another study, carried out by Grabovac, Guggi, and Bernkop-Schnürch established a ranking of the most studied polymers, showing that both thiolated chitosan and polycarbophil are the most mucoadhesive[34].

Currently, the addition of elements of sensitization and recognition continue being used for the design of polymers with more intelligent mechanisms of mucoadhesion. By binding functional groups within polymer chains, hydrogels can be made more sensitive to surrounding environmental conditions like temperature, moisture, pH, electrical fields and ionic forces.

Huang et al. (2000) proposed a mechanism in which units of the release system can specifically bind at the target surface. Certain amino acid sequences have complementary chains at mucous membrane and cellular surface. On contact with the mucous membrane, they can promote adhesion by binding to specific glycoproteins on this surface. Using this same mechanism, in the case of some diseases, changes occur in the glycoproteins, which can be attacked by complementary amino acid sequences linked to a release system, therefore increasing the affinity for diseased cells. The major problem with this strategy is finding the glycoproteins and their alterations in case of diseases.

With the advent of more intelligent mucoadhesive materials, it is possible to offer a unique carrying characteristic for many drugs. These can be designed for adhering onto any mucous membrane, for example ocular, buccal, respiratory, urinary, or gastrointestinal etc. Mucoadhesive materials can improve bioavailability, drug absorption and transport while reducing undesirable systemic effects. In summary, with these materials it is possible to develop novel systems for drugs currently used in therapy and to obtain new products at low cost[36].

Methods Of Analyzing Mucoadhesion

No technology has still been developed specifically to analyze mucoadhesion. Most of the tests available were adapted from other preexisting techniques but are useful and necessary for selecting the promising candidates as mucoadhesives as well as in elucidating their mechanisms of action.

In vitro and ex vivo tests

In vitro/ex vivo tests are important in the development of a controlled release bioadhesive system because they contribute to studies of permeation, release, compatibility, mechanical and physical stability, superficial interaction between formulation and mucous membrane and strength of the bioadhesive bond. These tests can simulate a number of administration routes including oral, buccal, periodontal, nasal, gastrointestinal, vaginal and rectal. The in vitro and ex vivo tests most prevalent in the literature are reported below.

Techniques utilizing gut sac of rats

The everted gut sac technique is an example of an ex vivo method. It has been used since 1954 to study intestinal transport. Santos et al. (1999) applied this method on mucoadhesion assays. It is easy to reproduce and can be performed in almost all laboratories. Figure 6 schematically represents the technique. A segment of intestinal tissue is removed from the rat, everted, and one of its ends sutured and filled with saline. The sacs are introduced into tubes containing the system under analysis at known concentrations, stirred, incubated and then removed. The percent adhesion rate of the release system onto the sac is determined by subtracting the residual mass from the initial mass[34] .

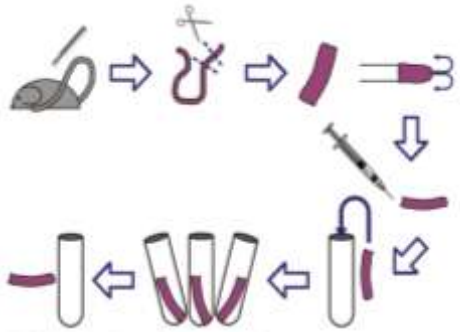

Fig :Everted gut sac procedure

Other techniques use non-everted gut sac. Takeuchi et al. (2005) filled rats' intestines with liposome suspensions. The sacs were sealed and incubated in saline. After a stipulated time, the number of liposomes adhered before (N0)and after (Ns) incubation was assessed with a coulter counter and the percent mucoadhesive was expressed by equation…

$$\%\,\textbf{adhesion} = \frac{N0-Ns}{N0} \times 100$$

The mucoadhesive effect of a system can also be evaluated by increases in gastrointestinal

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transit. Goto et al., 2006 incorporated fluorescent tracers into a system and quantified them by fluorescence spectroscopy in the stomach and intestinal mucus as a function of time[37].

Tests measuring mucoadhesive strength

Most in vitro/ex vivo methodologies found in the literature are based on the evaluation of mucoadhesive strength, that is, the force required to break the binding between the model membrane and the mucoadhesive.

Depending on the direction in which the mucoadhesive is separated from the substrate, is it possible to obtain the detachment, shear, and rupture tensile strengths.

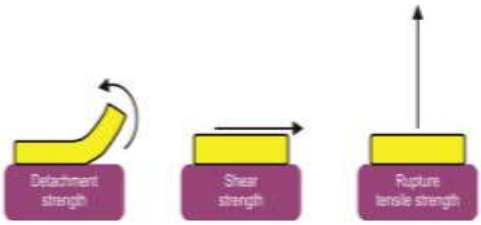

Fig: Different forces evaluated in mucoadhesion tests

The force most frequently evaluated in such tests is rupture tensile strength. Generally, the equipment used is a texture analyzer or a universal testing machine. In this test, the force required to remove the formulation from a model membrane is measured, which can be a disc composed of mucin, a piece of animal mucous membrane, generally porcine nasal mucus or intestinal mucus from rats . Based on results, a force-distance curve can be plotted which yields the force required to detach the mucin disc from the surface with the formulation , the tensile work (area under the curve during the detachment process), the peak force and the deformation to failure . This method is more frequently used to analyze solid systems like microspheres , although there are also studies on semi-solid materials[40].

Fig:Bioadhesion test using the texture analyzer

In addition to rupture tensile strength, the texture analyzer can also, as inferred by its name, evaluate the texture of the formulations and assess other mechanicalproperties of the system. A mobile arm containing an analytical probe forces down into a sample held in a flask placed on the equipment's platform. Speed rate, time and depth are preset. From the resulting force-time and forcedistance plots, it is possible to calculate the hardness (force required to reach a given deformation), compressibility (work required to deform the product during the compression), and adhesiveness (work required to overcome the attraction forces between the surfaces of sample and probe). Using this technique, it is possible to perform a previous evaluation of the material's adhesive capacity, evidencing mucoadhesion properties .

Mucoadhesion strength can also be measured in terms of shear strength. This test measures the force required to separate two parallel glass slides covered with the polymer and with a mucus film. This can also be done using Wilhemy's model, in which a glass plate is suspended by a microforce balance and immersed in a sample of mucus under controlled temperature. The force required to pull the plate out of the sample is then measured under constant experimental conditions. Although measures taken by this method are reproducible, the technique involves no biological tissue and therefore does not provide a realistic simulation of biological conditions.

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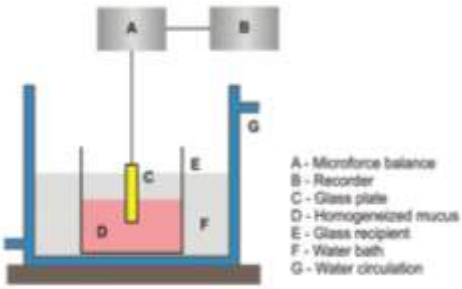

Fig :Apparatus to determine mucoadhesion in vitro, using Wilhemy's technique

Wilhemy's plate technique, or the microforce balance technique, can also be modified in order to measure the specific adhesion force of microparticles. This involves the use of a microtensiometer and a microforce balance and is specific, yielding both contact angle and surface tension. The mucous membrane is placed in a small mobile chamber with both pH and physiological temperature controlled. A unique microsphere is attached by a thread to the stationary microbalance.

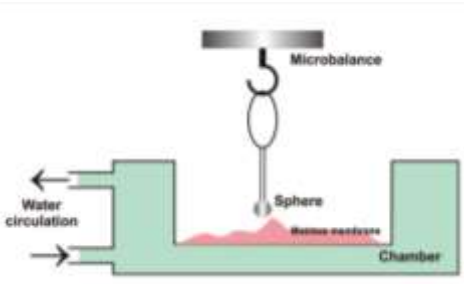

Fig:Microbalance method for measuring mucoadhesion.

The chamber with the mucous membrane is raised until it comes into contact with the microsphere and, after contact time, is lowered back to the initial position. Following the trajectory, and with the aid of software, results can be obtained for several parameters such as fracture strength, deformation and rupture tensile strength, from a load versus deformation curve[42].

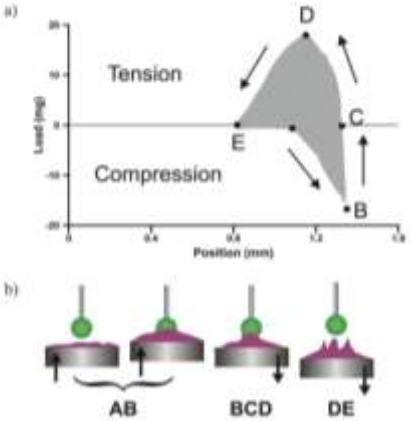

Fig: a) Typical load versus deformation curve; b) Progression of forces applied for corresponding graph

The microforce balance is not indicated for microspheres smaller than 300 μm, but has the advantage of simulating physiological conditions and providing results at a more microscopic level, besides being more reproducible and sensitive.

Rheological Methods

This category of methods are all carried out in vitro and were first proposed by Hassan and Gallo (1990), who used viscosimetric assays to macroscopically analyze the formulation-mucin interaction. From this test, it is possible to obtain the mucoadhesion force by monitoring the viscosimetric changes of the system constituted by the mixture of the polymer chosen and mucin. The energy of the physical and chemical bonds of the mucin-polymer interaction can be transformed into mechanical energy or work. This work, which causes the rearrangements of the macromolecules, is the basis of the change in viscosity. A way to analyze the coefficient of viscosity of a hydrophilic dispersion containing mucin plus the mucoadhesive polymer is through the contribution of each component, which results in equation

$\eta_t = \eta_m + \eta_p + \eta_b$ ..... [1]

where ηt is the coefficient of viscosity of the system, and ηm and ηp are the coefficients of viscosity of mucin and bioadhesive polymer, respectively. The bioadhesion component, ηb , can be obtained from equation, resulting in equation

$\eta_b = \eta_t - \eta_m - \eta_p$ .....[2]

For above both equations to be valid, all components should be measured at the same

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concentration, temperature, time and shear gradient. The bioadhesion force, F, is determined by equation 3

$F = \eta_b \, s$ ……...[3]

where σ is the shear gradient.

The main disadvantage of this method is the breakdown of the polymer and mucin network under continuous flow. To avoid this problem, the method was adapted using oscillatory rheology . Based on the same assumption that the rheological response of polymer-mucin mixture should be greater than the contributions from the gel and isolated mucin, a parameter called rheological synergism can be obtained. This method is more advantageous than the original, since oscillatory rheology is a non-destructive technique and simultaneously measures viscosity and elastic behavior and can be used to determine mucoadhesion between polymers and mucin[7] .

The evaluation of rheological synergism can be done through two types of oscillatory assays: stress sweep and frequency sweep.

In stress sweep, the elastic (G´) and viscous (G´´) moduli are obtained under constant frequency. This is used to investigate the influence of stress on the dynamic modulus, which should be obtained in the linear viscoelastic region, that is, the region where the material response is characteristic for its microstructure. Above this region, the structure is destroyed. The magnitude of the moduli is a qualitative indication of the system structure. Three situations can be found for polymeric dispersion: G'>> G" for a chemically interconnected system, G'>G" for chains with secondary bonds, and G'≤ G" for dispersions with physically-bound molecules[25]. The quantitative measure of rheological synergism (ΔG') can be calculated either in relation to G'or G" , as shown in equation 4

$\triangle G' = G_{mixtures} - [G'_{polymer} + G'_{mucin}]$ …[4]

In frequency sweep, stress is maintained constant. The structure of the system can remain intact during the assay if it is conducted in the linear viscoelastic region. Under constant stress and at low frequencies, better structured systems present greater elastic modulus than viscous modulus and both are independent of frequency. On a loglog graph, they are represented by a constant straight line. For less organized systems, dynamic moduli are dependent on the frequency and a slope is observed.

This test enables analysis of the dynamic viscoelastic parameters corresponding to the same frequency as a function of polymer or mucin concentration, yielding the rheological behavior in relation to the concentration of the system constituents.Hägerström (2003) reveals an alternative parameter of rheological synergism, called relative rheological synergism parameter (DG´relative), calculated from equation 5 and with which it is possible to quantitatively compare the force of polymer-mucin mixture with the isolated polymer

$$\Delta G'_{relative} = \frac{\Delta G'}{G'_p} = \frac{\left(G'_{mixture} - G'_p\right)}{G'_p} \quad [5]$$

where DG´ is the rheological synergism, given by the difference between elastic modulus of the mixture ($DG'_{mixture}$) and the elastic modulus of the polymer (G'_p).

However, $DG'_{relative}$ has the disadvantage of a negative limit up to -1, while the positive values run to infinity[11]. Therefore, the magnitude of positive values cannot be compared with that of negative values. Thus, a new relative parameter was proposed called the logarithmic relation of elastic module (log G´), which is given by the ratio between elastic modulus of the mixture (Gmix) and the elastic modulus of the polymer (G´p), as indicated in equation 6

$$\log G' = \log\left(\frac{G'_{mix}}{G'_p}\right) \quad [6]$$

This parameter offers the advantage that both positive and negative values have the same magnitude, and are therefore comparable. For instance, the value 1 means that G´ of the mixture is 10-fold greater than that of the isolated polymer.

Rheological tests are performed totally in vitro and consequently are conducted in combination with the rupture tensile strength test, most frequently used in studies on mucoadhesion. The experimental conditions of both tests differ and there are cases in which the techniques are complementary. Rheology measures the mechanical properties of the system, i.e., the resistance against flow and deformation, assessing the changes the system undergoes in the presence of mucin. However, rheology does not provide any direct information on what occurs at the interface, because the two phases – mucin and polymer – are mixed together prior to the experiment. In the rupture tensile strength test, the interface is artificially created. Even with this difference, when

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the mucin-polymer produces rheological synergism, a corresponding structure organization is observed at the mucoadhesive interface. The rupture tensile strength test can be applied to solids and semi-solids, while rheology is applicable to semi-solids and liquids. Experimental conditions are critical in the rupture tensile strength test and there are several variables (sample layer, hydration, time of hydration, sample load, time of loading, detachment rate, etc.), which should be optimized and set in order to produce reproducible results[36]. The reproducibility of rheological measures is reasonably good, since the measures are taken on already balanced mixtures; composition, pH, and temperature can be carefully controlled and therefore fewer repetitions are necessary to obtain statistically significant data. Thus, it can be concluded that both methods contribute to different extents toward explaining the mucoadhesive phenomenon, depending on the mucoadhesion mechanism involved, system type, polymer used, etc[39].

Tests analyzing molecular interactions involved in mucoadhesion

The general problem arising from methods that show the adhesion force and from the rheological methods is that the mucoadhesive response is seen macroscopically while the interactions occur at a microscopic level. The use of low frequency dielectric spectroscopy represents an attempt to study gel-mucus interactions near the molecular level. It evaluates the possible physicochemical interactions between molecules and glycoproteins of the mucus at the interface, which is considered the step preceding the formation of bonds during the mucoadhesion process. This technique involves the study of material response to the application of an electrical field. A sinusoidal voltage is applied throughout the sample and the response is measured in function of the frequency. From the responses, the impedance or permittivity of the sample is obtained and the property of charges changing in the system can be determined. This technique can provide information about the compatibility between mucus and mucoadhesive system by means of the evaluation of the movement of the charged particles. This compatibility is achieved according to the ease with which the particle crosses the barrier between the gel and mucous membrane. The dielectric measures reveal information about the gel and the mucous membrane separately, and about the interface between them[9].

Since the mucoadhesion process can be a consequence of interactions between the mucus layer and the mucoadhesive polymer, it is highly dependent upon the molecular structure, including its charge. It is also well known that glycoproteins molecules, which form the mucus structure, are negative at physiological pH. By means of zeta potential, it is possible to understand the polymer-mucin electrostatic interactions (Takeuchi et al., 2005). The zeta potential of dispersion is defined as the potential between the liquid superficial layer surrounding the dispersed particle and the remaining solution volume. It is a measure of the net surface charge of particles in a dispersed system. In this test, the mucin particles are suspended in an appropriate buffer and mixed with a solution of the polymer. If the addition of the polymer changes the zeta potential value of the mucin particles, this can suggest greater affinity between polymer and mucin particles[11].

Another technique being applied to evaluate molecular interactions is the optical biosensor, or resonant mirror biosensor technique. Sigurdsson, Loftsson and Lehr (2006) used this technique to measure the interactionbetween glycoproteins of the mucus and different polymers. It allows the monitoring of any interaction between two unknown molecules in real time, since one of them can be immobilized with covalent or non-covalent on the system surface while the other remains in solution at the surface. The molecules in solution, when binding to the immobilized molecules, alter the refraction index of the medium and this change is detected by the screening of a laser beam. The results of this study suggested the need for a clearer definition of mucoadhesion, because they called into question the polymers that are swelling dependent and undergo in situ gelification, because they do not seem to interact with glycoproteins, although they are called mucoadhesives[6].

Another test using the same principle, the Biacore test, was applied for the analysis of mucoadhesion by Takeuchi et al. (2005). This test is based on the passage of a mucin suspension through a sensor containing the immobilized polymer. When a mucin particle binds to the polymer at the sensor, both the solute concentration and the refraction index on this surface undergo changes, where the interaction is quantitatively evaluated and reproduced on a diagram. The sensor is a chip with a glass surface covered in a fine gold layer, where functional groups are introduced and the polymer is attached[21].

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Imaging Methods

Optical microscopes offer insufficient resolution for studying effects at a molecular level. For such investigations, a resolution at micro- or nanometric level is needed. Electronic microscopy gives a larger view, but the environmental conditions in which the sample must be submitted are far from the physiological conditions. For instance, the samples are analyzed in a vacuum chamber and generally are covered with a metallic film to avoid changes caused by the electronic rays[19].

Atomic force microscopy (AFM) is a relatively new technique that overcomes such restrictions, because it can be used under any environmental conditions, in air, liquids or vacuum. It enlarges more than 109 -fold, which enables visualization of isolated atoms and offers a tridimensional image of the surface. The equipment has a support combined with a probe perpendicularly attached to it. This tip moves toward a plane parallel to the sample, acquiring its topographic characteristics and the tip position is recorded by an optic deflection system: a laser beam is reflected onto the support and its position is then further reflected by a mirror reaching a photodiode sensor. A force-distance curve is plotted to measure the forces between this tip and the surface of interest. This curve is then used in bioadhesion studies. This entails coating the tip in adhesive material, which is generally spherical in shape and then the interaction with the surface, in this case the mucous membrane, can be measured[23] .

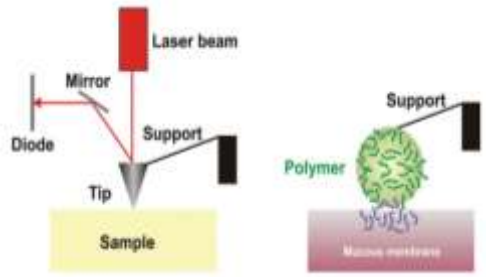

Fig: Constituents of AFM and the adaptations made for measuring the adhesive force between polymer and mucus surface. Adapted from Cleary, Bromberg and Magner, (2004), Mathiowitz, Chickering and Lehr (1999).

Besides AFM, there are other techniques using photographic images, such as fluorescence microscopy and confocal laser scanning microscopy (CSLM). Results achieved in ex vivo tests like the non-everted gut sac test, can be better visualized with this technique. Using radioisotopes or radioactive markers, it is possible to trace the polymer or the substance to be incorporated into the release system, where their location is visualized on the specific microscope, after the excision of the membrane. Takeuchi et al. (2005) used CSLM to analyze liposomes formulated with a fluorescent tracer and administered by the oral route in rats. The intestines were removed at an appropriate time after administration and the retention of the formulation was verified through the images achieved on the confocal microscope[1].

In the specific case of bioadhesive microspheres, the greater difficulty in their development is the sensitive quantification of the bioadhesive interactions under physiological conditions. Several techniques are being developed to measure the adhesion of great volumes in this kind of sample and others to offer more qualitative data. The previously described microforce balance methodology was an attempt to circumvent this difficulty. In parallel, another technology was developed, Electromagnetic Force-transduction (EFT). In addition to information about bioadhesive forces, this technology also offers the simultaneous video image of the interactions, with high resolution and under physiological conditions. The mucous membrane is mounted in a compartment under physiological conditions and the microsphere is positioned directly below the magnetic probe. The compartment is slowly moved down, in an opposite direction to the probe, and the video camera is used to detect sphere movement. According to the movement, the control system increases the magnetic current and the resulting magnetic force (Fm) pulls the sphere to its initial position, separating it from the tissue. After the experiment, the magnetic current is converted into force and the computer calculates the parameters of adhesion. The mucous membrane to be analyzed can be attained after an experiment using an everted gut sac[33].

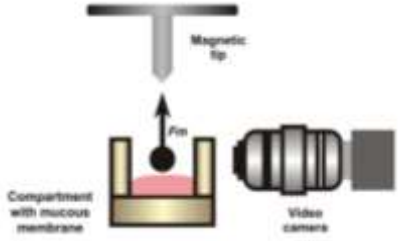

Fig: Elements of EFT.Adapted from Mathiowitz, Chickering and Lehr (1999).

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An alternative technique which also uses a video camera is the flow-channel method. A fine glass channel is filled with an aqueous bovine submaxillary mucin solution maintained at 37 ºC and humid air is passed through the channel. A particle of the bioadhesive polymer is placed in the mucin gel and both the static and dynamic behaviors are monitored by the camera at frequent time intervals[3] .

Falling Liquid Film Method

Nielsen, Schubert and Hansen (1998) used a method proposed by Rango Rao and Buri (1989) in which the chosen mucous membrane is placed in a stainless steel cylindrical tube, which has been longitudinally cut. This support is placed inclined in a cylindrical cell with a temperature controlled at 37 ºC. An isotonic solution is pumped through the mucous membrane and collected in a beaker. Subsequently, in the case of particulate systems, the amount remaining on the mucous membrane can be counted with the aid of a coulter counter. For semi-solid systems, the non adhered mucoadhesive can be quantified by high performance liquid chromatography (Nielsen, Schubert, Hansen, 1998). In this later case, porcine stomach, intestinal and buccal mucus were tested, and also jejunum from rabbits. The validation of this method showed that the type of mucus used does not influence the results. The release systems tested were precursors of liquid crystals constituted by monoglycerides. This methodology allows the visualization of formation of liquid-crystalline mesophase on the mucous membrane after the flowing of the fluids and through analysis by means of polarized light microscopy[41].

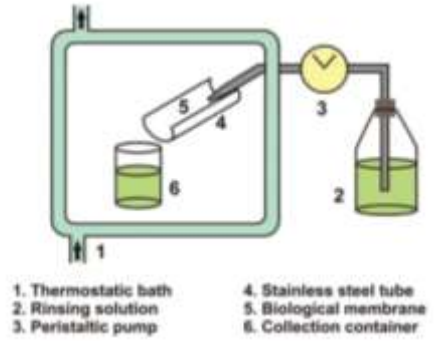

Fig: Schematic representation of in vitro model used by Nielsen, Schubert and Hansen (1998), adapted from Rango Rao and Buri (1989).

In vivo tests

There is scant information available on the in vivo behavior of mucoadhesive formulations, especially in humans. Säkkinen et al. (2006) applied gamma scintigraphy to analyze mucoadhesion in vivo of chitosan within the gastrointestinal tract. Gamma scintigraphy allows the immediate visualization of all the formulation transit, with low exposure of the subjects to radiation. The study emphasized the importance of in vivo studies, because although chitosan exhibits an outstanding mucoadhesion capacity in vitro, the retention time at the absorption site in the human gastrointestinal tract was relatively short and not sufficiently reproducible. The gastrointestinal transit time in animals can also be evaluated in a non-invasive way, in which the release systems can be formulated with opaque radioisotopes and signals can be followed by X-rays, without affecting normal gastrointestinal motility[24].

The number of methodologies applied to analyze mucoadhesion is constantly growing, although the useof different methods may sometimes lead to incoherence among results due to the heterogeneity of parameters and conditions used . Ahuja et al. (1997) examined various studies that used the tension resistance method and each had employed different models of mucous membrane and equipment. Despite the large body of evidence obtained to date, further investigations aimed at standardizing the methodologies are warranted[30].

Evaluation parameters :

a. Bulk density for powder:

Calculated based on following formula
Bulk density ($\rho 0$) = M/VO
Where,
M = mass of powder taken
V0= apparent untapped volume

b. Tapped density:

Calculated based on following formula
Tapped density (ρt) = M/Vf
Where,
M = Weight of sample powder taken
V f = Tapped volume

c. Hardness:

he hardness of five tablets was measured using Pfizer hardness tester. It is expressed in kg/cm2.

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d. Thickness and diameter:
Thickness and diameter of the prepared tablets were evaluated with the help of vernier calipers and screw gauge.

e. Friability: The friability of the tablets was determined using Roche friabilator. 20 tablets were initially weighed and transferred into the friabilator. The friabilator was operated at 25 rpm for 4 min. After 4 min the tablets were weighed again. The friability was then calculated using the formula[12],

$$\text{Friability (\%)} = \frac{\text{Initial weight} - \text{final weight}}{\text{Initial weight}} * 100$$

f. Weight variation:
Twenty tablets were randomly selected from each batch and individually weighed. The average weight and standard deviation of 20 tablets was calculated. The batch passes the test for weight variation test if not more than two of the individual tablet weights deviate from the average weight by more than the percentage shown in table 9 and none deviate by more than twice the percentage ± 7.5[13].

g. Drug content estimation:
Three tablets were crushed into powder, the quantity of powder equivalent to average weight of formulation was weighed and taken in a volumetric flask dissolved in 15 ml of methanol, the solution is filtered through whatman filterpaper, from this 1 ml of solution is withdrawn and diluted to 10 ml. Again from this, 1 ml of solution is withdrawn and diluted to 10 ml, absorbance is taken at 250 nm and % drug content is calculated by the formula**,**

$$\text{Drug content} = \frac{\text{Absorbance}}{1} \times \text{Dose} \times \text{Dilution factor} \times \text{Slope}\ 1000$$

$$\text{Drug content \%} = \frac{\text{Drug Content}}{\text{Dose of the formulation}} \times 100$$ [15]

h. % swelling study:
Buccal tablets were weighed individually (W1) and placed separately in 2 % agar gel plates with the core facing the gel surface and incubated at 37 ± 0.1°C. The tablets were removed from the petridish and excess surface water removed carefully using filter paper[41]. The swollen tablet was then reweighed (W2) and the swelling index was calculated using following formula. 27

$$\text{Swelling index} = \frac{\text{Final weight (W2)} - \text{Initial weight (W1)}}{\text{Initial weight (W1)}} \times 100$$

i. Matrix erosion: Tablets initial weight w

Matrix erosion: Tablets initial weight was noted down (W1). Swollen tablets were dried at 60 °C for 24 h in an oven and kept in desecatorfor 48 h and reweighed (W3). % matrix erosion were calculated using following formula, 21 W1-W3 % Matrix erosion= × 100 W3

i.Surface pH study:
Surface pH studies were carried out in order to investigate the possibility of any side effects. This has to be studied as the alkaline or acidic pH irritates buccal mucosa. The tablet was allowed to swell by keeping in contact with 1ml distilled water in a petridish for 2 h at room temperature. The pH was identified by bringing the electrode into contact with tablet surface and allowing the surface to equilibrate for 1 min[33].

j. Ex-vivo mucoadhesive time:
The Ex-vivo mucoadhesion time was examined after application of the buccal tablet on freshly cut sheep buccal mucosa. The fresh sheep buccal mucosa was tied on the glass slide, and a mucoadhesive core side of each tablet was wetted with 1 drop of phosphate buffer pH 6.8 and pasted to the sheep buccal mucosa by applying a light force with a fingertip for 30 seconds. The glass slide was then put in the beaker, which was filled with 200 mL of the phosphate buffer pH 6.8 and kept at 37 ± 1ºC. After 2 min, a slow stirring rate was applied to simulate the buccal cavity environment, and tablet adhesion was monitored for 12 h. The time for the tablet to detach from the sheep buccal mucosa was recorded as the mucoadhesion time[11].

Stability studies:
Stability studies for 2 months were carried out for the best formulation; the best formulation is kept under two different conditions like at 30 ± 2°C & 65 ± 5 % RH and other at 40 ± 2°C & 75 ± 5 % RH. After 30 days first month stability studies were carried out for important parameters like dissolution, diffusion, swelling index, matrix erosion, mucoadhesive strength, diameter, thickness, drug content. The same study is repeated after completion of 60 days[21].

Factors Affecting Mucoadhesion
Molecular weight
The mucoadhesive strength of a polymer increases with molecular weights above 100,000. Direct correlation between the mucoadhesive strength of polyoxyethylene polymers and their

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molecular weights lies in the range of 200,000–7,000,000[22]

Flexibility

Mucoadhesion starts with the diffusion of the polymer chains in the interfacial region. Therefore, it is important that the polymer chains contain a substantial degree of flexibility in order to achieve the desired entanglement with the mucus. The increased chain interpenetration was attributed to the increased structural flexibility of the polymer upon incorporation of polyethylene glycol. In general, mobility and flexibility of polymers can be related to their viscosities and diffusion coefficients, as higher flexibility of a polymer causes greater diffusion into the mucus network[23].

Cross-linking density

The average pore size, the number and average molecular weight of the cross-linked polymers, and the density of cross-linking are three important and inter-related structural parameters of a polymer network. Therefore, it seems reasonable that with increasing density of crosslinking, diffusion of water into the polymer network occurs at a lower rate which, in turn, causes an insufficient swelling of the polymer and a decreased rate of interpenetration between polymer and mucin[32].

Hydrogen bonding capacity

Hydrogen bonding is another important factor in mucoadhesion of a polymer. Desired polymers must have functional groups that are able to form hydrogen bonds, and flexibility of the polymer is important to improve this hydrogen bonding potential. Polymers such as poly(vinyl alcohol), hydroxylated methacrylate, and poly(methacrylic acid), as well as all their copolymers, have good hydrogen bonding capacity[12].

Hydration

Hydration is required for a mucoadhesive polymer to expand and create a proper macromolecular mes of sufficient size, and also to induce mobility in the polymer chains in order to enhance the interpenetration process between polymer and mucin. Polymer swelling permits a mechanical entanglement by exposing the bioadhesive sites for hydrogen bonding and/or electrostatic interaction between the polymer and the mucus network.However, a critical degree of hydration of the mucoadhesive polymer exists where optimum swelling and mucoadhesion occurs[13].

Charge

Some generalizations about the charge of bioadhesive polymers have been made previously, where nonionic polymers appear to undergo a smaller degree of adhesion compared to anionic polymers. Strong anionic charge on the polymer is one of the required characteristics for mucoadhesion.Some cationic polymers are likely to demonstrate superior mucoadhesive properties, especially in a neutral or slightly alkaline medium. Additionally, some cationic high–molecular-weight polymers, such as chitosan, have shown to possess good adhesive properties.is no significant literature about the influence of the charge of the membrane on the mucoadhesion but the pH of the membrane affects the mucoadhesion as it can influence the ionized or un-ionized forms of the polymers[23].

Concentration

The importance of this factor lies in the development of a strong adhesive bond with the mucus, and can be explained by the polymer chain length available for penetration into the mucus layer. When the concentration of the polymer is too low, the number of penetrating polymer chains per unit volume of the mucus is small and the interaction between polymer and mucus is unstable. In general, the more concentrated polymer would result in a longer penetrating chain length and better adhesion. However, for each polymer, there is a critical concentration, above which the polymer produces an “unperturbed” state due to a significantly coiled structure. As a result, the accessibility of the solvent to the polymer decreases, and chain penetration of the polymer is drastically reduced. Therefore, higher concentrations of polymers do not necessarily improve and, in some cases, actually diminish mucoadhesive properties.One of the studies addressing this factor demonstrated that high concentrations of flexible polymeric films based on polyvinylpyrrolidone or poly(vinyl alcohol) as film-forming polymers did not further enhance the mucoadhesive properties of the polymer[33].

Sites for Mucoadhesive Drug Delivery Systems

The common sites of application where mucoadhesive polymers have the ability to deliver pharmacologically active agents include oral cavity, eye conjunctiva, vagina, nasal cavity and GIT.

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The buccal cavity has a very limited surface area of around 50 cm^2 but the easy access to the site makes it a preferred location for delivering active agents. The site provides an opportunity to deliver pharmacologically active agents systemically by avoiding hepatic first-pass metabolism in addition to the local treatment of the oral lesions.

The sublingual mucosa is relatively more permeable than the buccal mucosa due to the presence of large number of smooth muscle and immobile mucosa. Hence, formulations for sublingual delivery are designed to release the active agent quickly while mucoadhesive formulation is of importance for the delivery of active agents to the buccal mucosa, where the active agent has to be released in a controlled manner. This makes the buccal cavity more suitable for mucoadhesive drug delivery.[18] The various mucoadhesive polymers used for the development of buccal delivery systems include cyanoacrylates, polyacrylic acid, sodium carboxymethylcellulose, hyaluronic acid, hydroxypropylcellulose, polycarbophil, chitosan and gellan. The delivery systems are generally coated with a drug and water impermeable film so as to prevent the washing of the active agent by the saliva[40].

Like buccal cavity, nasal cavity also provides a potential site for the development of formulations where mucoadhesive polymers can play an important role. The nasal mucosal layer has a surface area of around 150–200 cm2 . The residence time of a particulate matter in the nasal mucosa varies between 15 and 30 min, which has been attributed to the increased activity of the mucociliary layer in the presence of foreign particulate matter. The polymers used in the development of formulations for the development of nasal delivery system include copolymer of methyl vinyl ether, hydroxypropylmethylcellulose (HPMC), sodium carboxymethylcellulose, carbopol-934P and Eudragit RL100[15].

Due to the continuous formation of tears and blinking of eye lids, there is a rapid removal of the active medicament from the ocular cavity, which results in the poor bioavailability of the active agents. This can be minimized by delivering the drugs using ocular insert or patches. The mucoadhesive polymers used for the ocular delivery include thiolated poly(acrylic acid), poloxamer, celluloseacetophthalate, methyl cellulose, hydroxy ethyl cellulose, poly(amidoamine) dendrimers, poly(dimethyl siloxane) and poly(vinyl pyrrolidone)[25].

The vaginal and the rectal lumen have also been explored for the delivery of the active agents both systemically and locally. The active agents meant for the systemic delivery by this route of administration bypass the hepatic first-pass metabolism. Quite often, the delivery systems suffer from migration within the vaginal/rectal lumen, which might affect the delivery of the active agent to the specific location. The use of mucoadhesive polymers for the development of delivery system helps in reducing the migration of the same, thereby promoting better therapeutic efficacy. The polymers used in the development of vaginal and rectal delivery systems include mucin, gelatin, polycarbophil and poloxamer.

GIT is also a potential site which has been explored for a long time for the development of mucoadhesive based formulations. The modulation of the transit time of the delivery systems in a particular location of the gastrointestinal system by using mucoadhesive polymers has generated much interest among researchers around the world. The various mucoadhesive polymers which have been used for the development of oral delivery systems include chitosan, poly(acrylic acid), alginate, poly(methacrylic acid) and sodium carboxymethyl cellulose.

Each site of mucoadhesion has its own advantages and disadvantages along with the basic property of prolonged residence of dosage form at that particular site. In buccal and sublingual sites, there is an advantage of fast onset along with bypassing the first-pass metabolism, but these sites suffer from inconvenience because of taste and intake of food. In GIT, there is a chance for improved amount of absorption because of microvilli, but it has a drawback of acid instability and first-pass effects. Rectal and vaginal sites are the best ones for the local action of the drug but they suffer from inconvenience of administration. Nasal and ophthalmic routes have another drawback of mucociliary drainage that would clear the dosage form from the site[35].

Mucoadhesive Dosage Forms

Tablets

Tablets are small, flat, and oval, with a diameter of approximately 5–8 mm.[28] Unlike the conventional tablets, mucoadhesive tablets allow for drinking and speaking without major discomfort. They soften, adhere to the mucosa, and are retained in position until dissolution and/ or release is complete. Mucoadhesive tablets, in general, have the potential to be used for controlled

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release drug delivery, but coupling of mucoadhesive properties to tablet has additional advantages, for example, it offers efficient absorption and enhanced bioavailability of the drugs due to a high surface to volume ratio and facilitates a much more intimate contact with the mucus layer. Mucoadhesive tablets can be tailored to adhere to any mucosal tissue including those found in stomach, thus offering the possibilities of localized as well as systemic controlled release of drugs. The application of mucoadhesive tablets to the mucosal tissues of gastric epithelium is used for administration of drugs for localized action. Mucoadhesive tablets are widely used because they release the drug for a prolonged period, reduce frequency of drug administration and improve the patient compliance. The major drawback of mucoadhesive tablets is their lack of physical flexibility, leading to poor patient compliance for long-term and repeated use.

Films

Mucoadhesive films may be preferred over adhesive tablets in terms of flexibility and comfort. In addition, they can circumvent the relatively short residence time of oral gels on the mucosa, which are easily washed away and removed by saliva. Moreover, in the case of local delivery for oral diseases, the films also help protect the wound surface, thus helping to reduce pain, and treat the disease more effectively. An ideal film should be flexible, elastic, and soft, yet adequately strong to withstand breakage due to stress from mouth movements. It must also possess good mucoadhesive strength in order to be retained in the mouth for the desired duration of action. Swelling of film, if it occurs, should not be too extensive in order to prevent discomfort[11].

Patches

Patches are laminates consisting of an impermeable backing layer, a drug-containing reservoir layer from which the drug is released in a controlled manner, and a mucoadhesive surface for mucosal attachment. Patch systems are similar to those used in transdermal drug delivery. Two methods used to prepare adhesive patches include solvent casting and direct milling. In the solvent casting method, the intermediate sheet from which patches are punched is prepared by casting the solution of the drug and polymer(s) onto a backing layer sheet, and subsequently allowing the solvent(s) to evaporate. In the direct milling method, formulation constituents are homogeneously mixed and compressed to the desired thickness, and patches of predetermined size and shape are then cut or punched out. An impermeable backing layer may also be applied to control the direction of drug release, prevent drug loss, and minimize deformation and disintegration of the device during the application period[11].

Gels and ointments

Semisolid dosage forms, such as gels and ointments, have the advantage of easy dispersion throughout the oral mucosa. However, drug dosing from semisolid dosage forms may not be as accurate as from tablets, patches, or films. Poor retention of the gels at the site of application has been overcome by using mucoadhesive formulations. Certain mucoadhesive polymers, for example, sodium carboxymethylcellulose,[35] carbopol,[36] hyaluronic acid,[37] and xanthan gum,[38] undergo a phase change from liquid to semisolid. This change enhances the viscosity, which results in sustained and controlled release of drugs. Hydrogels are also a promising dosage form for buccal drug delivery. They are formed from polymers that are hydrated in an aqueous environment and physically entrap drug molecules for subsequent slow release by diffusion or erosion.[39] The application of mucoadhesive gels provides an extended retention time in the oral cavity, adequate drug penetration, as well as high efficacy and patient acceptability. A major application of adhesive gels is the local delivery of medicinal agents for the treatment of periodontitis, which is an inflammatory and infectious disease that causes formation of pockets between the gum and the tooth, and can eventually cause loss of teeth. It has been suggested that mucoadhesive polymers might be useful for periodontitis therapy when incorporated in antimicrobial-containing formulations that are easily introduced into the periodontal pocket with a syringe.HPMC has been used as an adhesive ointment ingredient. Additionally, a highly viscous gel was developed from carbopal and hydroxypropylcellulose for ointment dosage forms that could be maintained on the tissue for up to 8 hours[11].

II. CONCLUSION :

Studies on mucoadhesive systems have focused on a broad array of aspects. It is a growth area whose goal is the development of new devices and more “intelligent” polymers, as well as the creation of new methodologies that can better elucidate the mucoadhesion phenomenon. With the

great influx of new molecules stemming from drug research, mucoadhesive systems may play an increasing role in the development of new pharmaceuticals.

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